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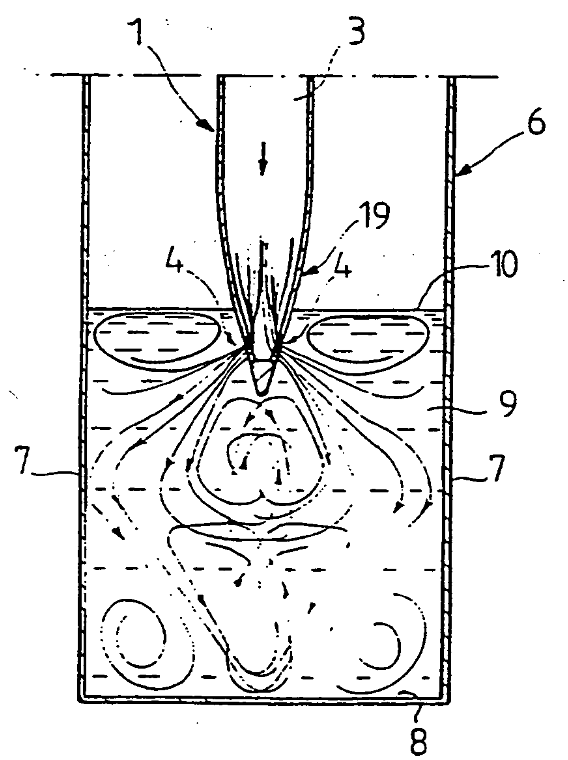
(57) Abstract

The present invention relates to a method for mixing of a liquid quantity for an analysis, and a mixing and measuring needle (1) which is suited for it. According to the invention for the mixing a needle (1) is used comprising a flowing duct (3) having at its end at least one aspiration and discharge opening (4), and the mixing is carried out by keeping the opening in a cuvette (6) or other such mixing container below the liquid surface (10) and by carrying out the mixing with a to-and-fro motion, wherein liquid is alternately aspirated from the container to a flowing duct of the needle and squirted back from the flowing duct into the container. The essence of the invention is that the flowing duct (3) has a necking (19) extending essentially to the opening (4) at the end of the duct, in the length of which duct the flow resistance of the duct is growing at least to tenfold, wherein at discharge stages from the opening according to the vena contracta phenomenon an at first convergent and thereafter turbulently divergent liquid flow (20) is discharged. The mixing can be combined with the measuring taking place in the needle simultaneously, wherein the needle is gradually emptied of the liquid to be measured. According to the invention the point part of the needle (1) is conical and has at least one opening (4) which has been directed aside from the longitudinal axis of the needle. The needle (1) can be manufactured by electroforming.



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Method for mixing a quantity of liquid in a container for an analysis, a mixing and measuring needle and method for manufacturing the needle

The invention relates to a method for mixing a quantity of liquid in a container for an analysis using a needle comprising a flowing duct having at its end at least one aspiration and discharge opening, by keeping the opening in a container below the liquid surface and by carrying the mixing with a to-and-fro motion, wherein the liquid is alternately aspirated from the container to a flowing duct of the needle and sprayed from the flowing duct back into the container. The method is especially intended for the mixing of liquid samples to be analyzed in measuring cuvettes of automatic analyzers in the field of clinical chemistry.

The components of the liquid quantity to be mixed in the cuvette are typically measured with a measuring needle, which is flexibly connected to an automatic microsyringe. The measuring is carried out so that a sample and a diluent or a reagent are aspirated with the syringe to the needle and optionally air bubbles to separate these from each other. The point of the measuring needle is moved to the cuvette, wherein the sample with its diluent or reagent is emptied. Thereafter the measured components and the reagents which optionally were in the cuvette in advance are mixed with each other. The mixing has been carried out mechanically e.g. by moving the point of the measuring needle in the cuvette by using a separate mixer or by carrying out the mixing with ultrasound. After the mixing the cuvette is transported to the incubation, measuring or other such further handling.

The mentioned known methods for mixing a liquid quantity all are more or less slow and tediuos. The use of a separate

mechanical mixer is tedious and comprises an extra step in the preparation of the liquid quantity for an analysis. The point of the measuring needle in turn is inefficient as a mechanical mixer because of its thinness.

From the published German Application DE 27 22 586 is known the in the beginning mentioned way of mixing, which is based on the to-and-fro movements. Because of the form of the needle the mixing is, anyhow, inefficient and it isn't mentioned for analytical purposes.

The purpose of this invention is to provide a solution, by which the liquid quantity to be analyzed can be mixed more efficiently than earlier without any need for a separate, moving mixer in the container. The invention is characterized in that a needle is used, in the flowing duct of which is a necking extending essentially to the opening at the end of the duct, in the length of which necking the flowing resistance of the duct is growing at least to tenfold, wherein at the discharge stages from the opening according to the vena contracta phenomenon an at first convergent and thereafter turbulently divergent liquid flow is discharged, and that the liquid quantity moving in the to-and-fro movements is at most about 20% of the total volume of the liquid quantity to be mixed.

According to the invention, the liquid quantity is mixed with the needle which can also be used for measuring of the components of the liquid quantity to the container. As distinct from the earlier known method there is, anyhow, no need to move the needle, but the mixing is based exclusively on the movements of the liquid provided by the needle. The conventional measuring wherein the liquid contained in a straight, constant diameter measuring needle is sprayed all at once to the container, is usually not adequate to cause any remarkable mixing of the liquid in the container, whereas with the to-and-fro motion of the rather small

liquid quantity mixing is taking place e.g. because of the fact that the liquid can be aspirated to the needle at a point near the opening at the end of its flowing duct and thereafter sprayed with pressure essentially rectilinearly to a point far away from said opening. Of the essence in the invention is that with help of the strong necking of the flowing duct and optionally additionally the suitable location of the one or several aspiration and discharge openings the according to the vena contracta phenomenon discharged flow, which is to be sprayed to the container, is made to cause turbulence in the liquid, securing an effective mixing.

One preferable embodiment of the invention is characterized in that the mixing is combined with the simultaneous measuring of the liquid by the needle, wherein the liquid quantities aspirated from the container to the needle are in average smaller than the liquid quantities sprayed to the container from the needle. In other words, the liquid to be measured is little by little discharged to the container during the measuring and mixing process. Such a method is quicker than carrying out the measuring and mixing separately, in addition to which an advantage is that the liquid to be measured is continuously rinsing the flowing duct inside the needle and prevents its contamination.

It is, of course, also possible first to carry out the measuring with the needle, wherein the liquid to be measured is first discharged from the needle to the container, whereafter the liquid in the container is mixed with said to-and-fro mixing movements. In this case the liquid quantity which is to be aspirated to the needle and to be sprayed from it back to the container can be kept constant during the hole mixing event.

In the mixing of the liquid quantity according to the invention the liquid quantity to be moved to-and-fro can

according to the invention be about 0.1-5%, most preferably about 0.1-2% of the total volume of the liquid quantity to be mixed. The volume of the liquid quantities handled by clinical analyzers varies between 1-1000μl, and a typical liquid quantity to be moved in the mixing is of order 1μl. If with the mixing has been combined with the measuring of the liquid, the liquid quantity to be measured can be e.g. about 10% of the total volume of the liquid quantity.

The frequency of the to-and-fro mixing movements in the mixing can be between about 5-200Hz, preferably between 10-100Hz, depending on e.g. the softness of the material of the feed pipe connected to the needle or the air optionally contained in the pipe.

A frequency of 50Hz for instance can be provided simply with rectified mains voltage.

The mixing according to the invention can take about 0.1-2 seconds, preferably about 0.2-1 seconds. If the mixing takes e.g. 0.5 seconds, it means at a frequency of 50Hz 25 aspiration and discharge stages. By the known methods the mixing typically takes 2-4 seconds, to which also the duration of the measuring has to be added. As mentioned above, the invention makes also possible the measuring in frame of the time used for the mixing, thus maximizing the saving of time which is attained by the invention.

The to-and-fro mixing movements which the invention requires are preferably provided by the repeated compressions exerted on the pipe of a soft material which is as an extension to the needle, by the action of which the volume of the pipe is changed. The compression will reduce the volume of the pipe and forces liquid to get discharged from the needle to the container. Ceasing of the compression will correspondingly increase the volume of the pipe, bringing about a aspiration which is directed from the container towards the flowing

duct of the needle. The repeated compressions can be provided e.g. by bringing the pipe between the piston of a solenoid valve and the solid surface opposite to it and by moving the piston to-and-fro by means of a spring force and a magnetic force engaged by an electric current. Alternatively the repeated compressions can be provided with a fork formed by two piezo crystals, wherein the pipe is situated between two against each other bending ceramic, metal coated plates.

In order to provide the according to the mentioned vena contracta phenomenon an at first convergent and thereafter turbulently divergent liquid flow the necking inside the flowing duct is preferably conical, having an angle of taper which can vary between about 15-60°.

Said vena contracta phenomenon can be effected with the needle the flowing duct of which is ending rectilinearly in the discharge opening at the point of the needle, if in the duct is said necking, along the length of which the flow resistance is strongly growing. In relation to mixing even more efficient is a needle wherein the flowing duct is ending before the point of the needle in one or several discharge openings which are directed to the side of the longitudinal axis of the needle. The liquid flows can then be directed on the sides of the needle towards the side walls of the container and from them further towards the bottom of the container. As a result of this the kinetic energy of the liquid flows is damping due the turbulence generated in the cuvette and due to the collision of the flows to each other.

The present invention is also directed to a mixing and measuring needle comprising a tubular shaft and a flowing duct inside it ending before the point of the needle to at least one aspiration and discharge opening which is directed aside from the longitudinal axis of the needle. Additionally

the invention is directed to a method for manufacturing such a needle. The needle is especially suited to be used in the above described method for mixing of the liquid quantity, which method can also include the measuring of the liquid.

The present automatical analyzers of clinical chemisty typically comprise a measuring needle which is flexibly connected to the automatic microsyringe. The measuring is carried out so that the sample and a diluent or a reagent, and optionally air bubbles in order to separate these from each other are aspirated with the syringe to the measuring needle. The point of the measuring needle is moved to the cuvette, where the sample with its diluents or reagensts is discharged. Thereafter the the measured components and the reagents which optionally were in the cuvette in advance are mixed mechanically e.g. by moving the measuring tip in the cuvette, by using a separate mixer or by carrying out the mixing by ultrasound. After the mixing the cuvette is transported to the incubation, measuring or other such further handling.

In the mentioned method are involved some disadvantages which arise from the measuring needle which is generally made of commercial metal pipe. In the measuring the tip of the needle is getting wet from outside, wherein there is an extra drop of liquid left outside the needle, causing a little error of the measuring volume. The error can be reduced by reducing the diameter of the needlen or its point, but then the liquid transportations are slowered and the most important property of an automatic analyzer, the speed, is weakened. If the measuring tip is used as a mixer, the narrower point additionally requires a longer mixing time. Additionally, when the diameter of the tubular measuring needle is reduced, the strength of the needle is jeopardized. The outer diameter of the needle has to be at least about 0.5mm and the thickness of its wall has to be at least about 0.1mm in order to give sufficient strength to

it. Then the volume of the drop of liquid outside the point of the needle is typically 0.02-0.2μl and the deviation is about 0.02-0.03μl. When the length of the measuring needle is over 100mm, the transportation of the liquid through it will take several seconds.

The needle according to the present invention without the above mentioned disadvantages linked to the measuring needles of the present art, and which in addition to measuring is also providing an efficient mixing of the liquid, is characterized by that the tip part of the needle has been formed conically tapering and that in the flowing duct there is a necking extending to said opening, along whic necking the flow resistance of the duct is growing to at least tenfold, wherein at the discharge stages from the opening is according to the vena contracta phenomenon an at first convergent and thereafter turbulently divergent liquid flow.

Since the discharge opening(s) of the needle according to the invention has/have been directed aside from the longitudinal axis of the needle, it is possible to direct the discharging liquid flow with the needle towards the side wall of a conventional straight measuring cuvette and from there further towards the bottom of the cuvette. When there are several discharge openings, the flows directed towards the walls and bottom of the cuvette are made to finally collide against each other. The result of this and said vena contracta phenomenon is that the liquid flows loose their kinetic energy due to the turbulence generated in the cuvette. This turbulence is providing an effective mixing of the liquid, which mixing can then be further secured with to-and-fro mixing movements which have been described above. As an advantage is that the mixing by moving the measuring needle or with a separate mixer is unnecessary, wherein the measuring is essentially accelerated.

The needle according to the invention can be mounted essentially as such to the clinical analyzers in place of the presently used needles. As a result the operation of the expensive analyzers costing even several hundreds of thousands Fmk (several tens of thousands USD) is accelerated and the capacity is increased by up to 30% without any increase in costs. At the same time the quality of analyses is getting better thanks to quicker and more realiable mixing.

One preferable application of the needle according to the invention is characterized in that the point of the needle is solid. The solid point gives structural strength to needle, which in turn gives an opportunity to make the walls of the needle narrower. The flowing duct inside the needle can thus be made wider than ever and the measuring quicker, and more effective in mixing the liquid.

According to an other preferable embodiment of the needle of the invention the point part of the needle is tapering off conically at an angle between about 15-60°, most preferably about 15-30°. The diameter of the point is thereby preferably less than 0.3mm, most preferably 0.2-0.3mm. When such a needle is lifted out from the liquid, the capillary forces of the liquid are dragging the liquid film and possible drops on the outer surface of the point with an accelerating speed towards the liquid. Thereby the extra liquid quantity which is left to the point of the needle is minimally small and additionally very accurately reproducible. The drop of liquid which is left in the known tubular measuring needles is essentially larger, in addition to which the reproducibility of the drop would require the needle to be lifted from the liquid as slowly as possible. In practise one has been forced to make a compromises between the reproducibility and the time used for the measuring. The present invention in contrast to that makes possible a quick measuring while the contamination from one

measuring to an other is minimal.

The solid point of the needle further gives an opportunity to form the point part of the needle in the shape of a dagger, wherein it will penetrate the rubber or plastics cap of the sample tube without any danger of obstructing the discharge openings which are directed aside. In the analyzers of the present art the caps of the sample tubes have to be removed before the sampling which is tedious and due to the possibility of splashing of samples containing AIDS vira even dangerous to the operator. In contrast to that, the present invention makes it possible to take a sample into the needle and to pull away the needle without taking off the cap from the tube. In the needle having a shape of a dagger there is more extra liquid left on the outer surface than by a needle the point of which has been shaped to a tapering circular cone, but this is not causing any harm because the extra liquid is wiped off the point of the needle while it is exiting the tube through the cap.

The discharge opening(s) of the mixing and measuring needle according to the invention can be directed obliquely forward in relation to the longitudinal axis of the needle. The optimal angle varies depending e.g. on the form of the cuvette. It is possible that there is only one discharge opening, but most preferably there are two or several disposed at regular intervals on the different sides of the needle. Only one discharge opening requires in practise that the needle is positioned to the cuvette near its side wall and the discharge opening is directed towards the middle of the cuvette. When there are two or several disharge openings, the needle can be positioned to the middle of the cuvette, wherein liquid flows are discharged on different sides of the cuvette. A needle with two discharge openings is suitable for a cuvette with a rectangular cross section, a needle with three discharge openings for a cuvette with a round cross section, and a needle with four discharge

openings for a cuvette with a square shaped cross section.

The needle according the present invention can be lifted during the measuring so that the discharge opening(s) follow(s) the rising of the liquid surface in the container. If the openings are just beneath the liquid surface, an effective turbulent mixing of the hole liquid volume of the container can be effected.

The method according to the invention for manufacturing of the mixing and measuring needle is characterized in that the manufacturing is carried out by electroforming by precipitating electrolytically a layer of a metal around an elongate needlelike mould, by removing the mould from inside the metal layer so that a tubular needle of metal is obtained having a longitudinal duct inside it, and by drilling on the side of the needle at the end of the duct at least one opening through the metal layer as an discharge opening of the duct.

The needle is provided with a solid, conically tapering or the shape of a dagger having point by precipitating an excess of the metal round the point of the mould and the needle point formed after the precipitation stage is worked to the desired form, wherein the extra metal is removed.

The invention will be described in more detail with the following examples with reference to the appended figures, wherein

Fig. 1 is a longitudinal section of one, with two discharge openings provided, at its point conically tapering point part of a mixing and measuring needle according to the invention,

Fig. 2 presents the needle of Fig. 1 seen from the direction of its point,

Fig. 3 presents corresponding the Fig. 2 a needle with three discharge openings,

Fig. 4 presents corresponding the Fig. 2 a needle with four discharge openings,

Fig. 5 presents corresponding the Fig. 2 a needle with one curved discharge opening,

Fig. 6 presents a at its point dagger-shaped needle according to the invention, seen from the side and partly opened,

Fig. 7 is section VII-VII from Fig. 6,

Fig. 8 is section VIII-VIII from Fig. 7,

Fig. 9 is section IX-IX from Fig. 7,

Fig. 10 presents the measuring of a liquid to a measuring cuvette with the needle according to Figs. 1 and 2,

Fig. 11 presents the measuring of a liquid to a measuring cuvette with the needle according to Fig. 5,

Figs. 12-14 present the stepwise manufacturing of a needle by electroforming,

Fig. 15 presents the aspiration stage of the inventive mixing of a liquid quantity, wherein liquid is aspirated from a container to the flowing duct of a needle,

Fig. 16 presents the discharge stage of the mixing, wherein liquid is sprayed from the needle back into the container,

Figs. 17 and 18 present the aspiration and discharge stages of the mixing according to the invention, wherein the needle

used is different from the one presented in Figs. 15 and 16, and

Fig. 19 presents the provision of to-and-fro mixing movements by the repeated compressions directed to the hose which is as an extension of the needle.

In connection with the drawings, the mixing and measuring needles and their manufacturing are first explained in the following, and thereafter the mixing according to the invention, which can be carried out besides with the described needles according to the invention, also with the needle ending directly in an axial opening (Figs. 15 and 16), which needle itself is not an object of the present invention.

The mixing and measuring needle (1) according to Figs. 1 and 2 comprises an elongate, tubular shaft (2) which defines inside itself a flowing duct (3). The shaft (2) is preferably of ironless nickel cobalt and it can have on its inner surface a layer of a noble metal. The flowing duct (3) is ending in two discharge openings (4), which have been directed obiquely forward at an angle of about 45° in relation to the longitudinal axis of the needle. The form of the point part of the needle (1) is according to Fig. 1 a tapering circular cone, and the needle is ending in the solid point (5). The diameter of flowing duct (3) is typically between 0.7mm and 2.5mm, the thickness of the wall of shaft (2) is typically 3-10% of the diameter of duct (3), the diameter of the point (5) of the needle is preferably about 0.2-0.3mm and diameter of the discharge opening (4) is preferably about 0.1-0.4mm. Of essence is that the flow resistance is increasing at the tapering part of the cone inside the flowing duct (3) to at least tenfold.

In Fig. 3 can be seen, looking upwards, the mixing and measuring needle (1), which is different from that presented

in Figs. 1 and 2 only in that there are three discharge openings (4) and that they are disposed at angles of 120° on the different sides of the needle. In Fig. 4 is correspondingly seen the needle (1) with four discharge openings (4) which are disposed at angles of 90° on the different sides of the needle.

In Fig. 5 the needle (1) only has one curved discharge opening (4). Otherwise even this needle corresponds the one presented in Figs. 1 and 2.

The mixing and measuring needle (1) presented in Figs. 6-9, the point part of which has been formed in the shape of a dagger, corresponds in respect of the flowing duct (3) and the discharge openings (4) inside the shaft the needle presented in Fig. 1. Also the side profiles of the needles (1) are essentially the same (cf. Figs. 1 and 6). Only seen from the front the shape of a dagger needle differs due to its broader form (Fig. 7). The needle has sharp edges 2′ and it is characterized in the ability to penetrate the rubber or plastics cap of the sample tube so that the sample can be aspirated ti the needle without taking off the cap from the tube. The disposing to the side of the discharge openings (4) of the needle (1) protects them from getting obstructed when they are passing through the cap. The needle (1) has preferably a titanium nitride coating, wherein the durability of edges (2′) of the section is increasing.

Fig. 10 presents the measuring of a liquid with the needle (1) of Figs. 1 and 2 to a straight, flat-bottomed cuvette (6). The cross section of cuvette (6) can be square shaped or rectangular, or it can also be round. The jets of liquid discharged according vena contracta phenomenon from the discharge openings (4) of the needle (1) are directed towards the side walls (7) of the cuvette (6), from where they are bent further towards the bottom (8) of the cuvette, to a minor ectent towards the surface (10) of the liquid

(9). According to Fig. 10, the jets are scattering, colliding to each other and loose their kinetic energy to the generated turbulence which causes mixing of the liquid (9) in the cuvette. During the measuring the needle (1) can be liftet so that että its point and the discharge opening are continuously kept near the surface (10) of the liquid.

Fig. 11 presents the measuring of a liquid with the needle (1) according to Fig. 5 to the cuvette (6), the cross section of which is most suitably roud. The needle (1) is taken to the cuvette (6) near its side wall (7), and the liquid is let to discharge from the curved discharge opening (4) of the needle, which opening is directed towards the centre of the cuvette.

The manufacture of the mixing and measuring needle (1) according to the invention 1 is preferably carried out by using electroformation. Figs. 12, 13, and 14 present the different phases of the manufacture. In the manufacture an elongate, needlelike aluminum mould (11) is used, inside which the is a duct (13) which extends to the point (12) of the mould. On the mould (11) is precipitated electrolytically a layer (14) of nickel cobalt wherein the share of cobalt is on most suitably some few percents. The thickness of the nickel cobalt layer (14) is otherwise corresponding the wall thickness of the needle (1) to be manufactured, but around the point part of the mould (11) nickel cobalt is precipitated in an excess so that the expansion (15) seen in Fig. 12 is provided. The intention of this is to provide to the needle (1) a solid, conically tapering point (5).

After the precipitation step the expansion (15) surrounding the point (12) of the mould is tooled by first making to its point the bevellings (16) according to Fig. 13 at an angle of 45° in relation to the longitudinal axis of the mould (11). Thereafter the borings (17) perpendicular to the bevellings (16) are drilled extending to the internal duct

(13). The borings (17) the discharge openings (4) of the needle to be manufactured, which openings have been directed obiquely forward at an anfle of 45° in relation to the longitudinal axis of the needle.

At following stage, presented by Fig. 14, the rest of the expansion (15) precipitated around the point part of the mould (11) is removed, except the solid, conical point (18) of the needle (1) which is forming. According to Fig. 14, the conical point (18) has been tooled to form an angle of 30°. At this stage the needle formed of the cobalt nickel layer (14) is completely finished, except the aluminum mould (11) which still is inside the layer, which mould is removed by leading inside the mould through the duct (13) and borings (17) hot lye which is dissolving the aluminum but doesn't do any harm to cobalt nickel. The final result is the mixing and measuring needle (1) of Fig. 1, where an electrolytically precipitated and tooled layer (14) of cobalt nickel forms the tubular shaft (2) of the needle, and the drilled borings (17) form the discharge openings (4) of the needle.

Also the needle (1) according to Figs. 6-9 having a dagger shaped point part can be manufactured by electroforming by using a completely similar aluminum mould (11). The only difference compared to the above presented is that the expansion (15) is not tooled to a tapering circular cone but to the dagger form according to Figs. 6 and 7.

The manufacturing process can, if desired, be modified so that a thin layer of a noble metal is first precipitated around the aluminum mould (11), and the nickel cobalt is thereafter precipitated on the noble metal layer. By this way an inner surface of a noble metal defining the flowing duct (3) is provided to the needle. Correspondingly, also the outer surface of the needle can be coated with a noble metal. It is also possible that the removal of the aluminum

mould is taking place already at an earlier stage than mentioned above, e.g. immediately after drilling of the borings (17).

In addition to electroformation, chemical deposition, sputtering and other methods known <u>per se</u> come into question in the manufacturing of the needle.

The mixing of the liquid quantity (9) according to Figs. 15 and 16 is taking place in a container (6), which comprises a cuvette used in automatic analyzers. The mixing is carried out with the needle (1), which comprises a tubular shaft (2), a flowing duct (3) inside the shaft, and a aspiration and discharge opening (4) at the point of the needle, at the end of the flowing duct. Otherwise the flowing duct (3) having a constant diameter is ending to the conical narrowing (19) extending to the opening (4) at the point of the needle (1), during the lengt of which narrowing the diameter of the duct is converging from e.g. about 1mm to below 0.5mm, so that the flow resistance increases to at least tenfold.

The mixing is taking place according to Figs. 15 and 16 with to-and-fro movements, wherein with the needle (1), the point (4) of which is below the liquid surface (10) in the cuvette, liquid is alternately aspirated from the cuvette to the flowing duct (3) inside the needle and squirted liquid from the duct back to the cuvette. Then the linear velocity of the liquid jet flowing to the cuvette at the medium distance of the liquid to be mixed is multiple compared to the velocity of the liquid flowing to the needle at the same point. The liquid quantity to be moved to-and-fro is typically of order 1μl, which is suitably 0.1-5% of the total volume of the liquid quantity (9) which is in the cuvette and is to be mixed, although it can be even more. The aspiration of the liquid is taking place according to the arrows in Fig. 15 near the aspiration and discharge

opening (4) quite evenly from the different sides from the opening. In contrast to that, at the discharge stage according to Fig. 16 the liquid jet (20) moves tar away from the opening (4) towards the bottom (8) of the cuvette, from where it turns according to the arrows back towards the surface (10) of the liquid, providing the mixing in the whole volume of the liquid quantity (9). This mixing is essentially enhanced by the vena contracta phenomenon induced by the tapering narrowing (19) of the flowing duct (3), according to which phenomenon the liquid jet (20) discharged from the opening (4) is first converging and thereafter broadening and begins to break up as a turbulence because of the aspiration influencing on the edges of the jet.

According to Figs. 15 and 16, to cuvette (6) can be mixed a liquid quantity (9) which has been measured in advance by what so ever method. For the measuring the same needle (1) can anyhow be used with which the liquid quantity (9) is thereafter mixed according to the invention. Further, it is possible that the measuring of the liquid to the cuvette (6) and mixing are taking place simultaneously. The liquid to be measured is then in the flowing duct (3) of the needle (1), and a little larger liquid quantity is each time squirted from the needle to the cuvette (6) than is aspirated from the cuvette to the needle. Then the needle (1) is emptied from the liquid to be measured during the simultaneous measuring and mixing provided by the to-and-fro movements.

As an example of according to the invention of the simultaneous mixing and measuring the following can be presented: The cuvette contained 90μl of a liquid, which in order to test the mixing had been thickened with extra protein to 100-fold. To this, 10μl of an other liquid with a thickness corresponding that of water was measured with a needle, wherein the diameter of the two aspiration and discharge openings was 0,25 mm. The oparation took 1 second

and the frequency of the to-and-fro movements were 50Hz, wherein the operation comprized 50 successive aspiration and spraying stages. At each aspiration stage the liquid quantity aspirated to the needle was 1μl and the liquid quantity discarged at each discharge stage from the needle to the cuvette was 1.2μl. The linear flowing rate of the liquid jets discharged to the cuvette was about 5 meters per second as measured in air, because the mixing turbulence inside the liquid made the measuring impossible. The linear flowing rate of the aspiration stage as measured microscopically by using micropartikles is less than 0.01m/s. The mixing was optically complete at the end of the measuring, while the mixing by moving the needle with the speed of the moving method in question the mixing wasn't complete even with the mixing time of 5 second after the measuring.

The in Figs. 17 and 18 presented successive, repeated aspiration and discharge stages of the mixing of the liquid quantity (9) in the cuvette (6) correspond generally the above described ones. As a difference is anyhow the above described, to mixing used needle (3) according to Fig. 1, wherein the conically tapering flowing duct (3) ends before the point (5) of the needle to two aspiration and discharge openings (4) which have been directed obiquely forward in relation to the longitudinal direction of the needle. The point (5) of the needle (1) is sharply tapering and solid.

At the aspiration stage seen in Fig. 17 liquid is aspirated by the needle (1) from openings (4) to the flowing duct (3) inside the needle substantially in the same manner as with the needle according to Figs. 15 and 16. At the discharge stage according to Fig. 18 the liquid jets are directed from the openings (4) first towards the side walls (7) of the cuvette (6), from where they turn towards the bottom (8) of the cuvette and towards each other. At the same time there is a aquirting of the jets according to the above mentioned vena contracta phenomenon taking place, which is

contributing in the whole liquid volume (9) to generated, the liquid effectively mixing turbulence. The mixing is even more effective than in the case presented in Fig. 16, in addition to which one advantage is that the liquid jets directed towards the sides of the needle (1) better keep in place the liquid (9) to be mixed in the cuvette without any risk of splashing.

Fig. 19 represents needle (1), the point of which is in the liquid (9) to me mixed in the cuvette (6), which needle is connected to a feed pipe which is formed of a flexible rubber pipe or a flexible plastics or metal pipe (21). The pipe (21) has been led between a piezo crystal formed by two metal coated ceramic plates (22). By these bending plates (22) provided, on pipe (21) concentrated repeated compressions alternately reduce and add the volume of the pipe, so that the needle (1) when the pipe is largening is aspirating liquid from the cuvette (6) and when volume of the pipe is reduced, is squirting the liquid back to the cuvette. The plates (22) are most preferably at a little angle in relation to each other so that the compression seals the pipe (21) beginning from that end of the compression area which is more distant from the cuvette (6). This quarantees that the squirting is always directed towards the cuvette (6).

The corresponding to-and-fro movements of the liquid could also be provided by a solenoid valve by bringing the pipe (21) between a solid dolly and a piston which is engaged in turns to different directions by alternately a spring force and a magnetic force generated by a magnetic force.

It is evident to one skilled in the art that the different embodiments of the invention can also otherwise diverge from the facts presented above by the examples, within the patent claims. The mixing and measuring needle can for example in addition to the discharge openings comprise an axial

discharge opening at the point of the needle, from where the liquid flow is directed straight downwards.

Claims

1. Method for mixing a liquid quantity (9) in a container (6) for an analysis using a needle (1), which comprises a flowing duct (3), at the end of which is at least one aspiration and discharge opening (4), by keeping the opening in a container beneath the liquid surface (10) and carrying out mixing with a to-and-fro motion, wherein liquid is alternately aspirated from the container to a flowing duct in the needle and squirted from the flowing duct back into the container, **characterized** in that a needle (1) is used having in its flowing duct (3) a necking (19) extending essentially to the opening (4) at the end of the duct, in the length of which necking the flow resistance of the duct is growing at least to tenfold, wherein at the discharge stages from the opening according to the vena contracta phenomenon an at first convergent and thereafter turbulently divergent liquid flow (20) is discharged, and that the liquid quantity moving in the to-and-fro movements is at most about 20% of the total volume of the liquid quantity to be mixed.

2. Method according to claim 1, **characterized** in that the mixing has been connected with the simultaneous measuring carried out with the needle (1), wherein the liquid quantities aspirated from the container (6) to the needle are on an average smaller than the liquid quantities squirted from the needle to the container.

3. Method according to claim 1, **characterized** a measuring is first carried out with the needle (1), in which measuring the liquid to be measured is first discharged from the needle to the container (6), whereafter the liquid in the container (9) is mixed with said to-and-fro mixing movements.

4. Method according to any preceding claim, **characterized** in that the liquid quantity to be moved to-and-fro in the

mixing is about 0.1-5% of the total volume of the liquid quantity (9) to be mixed.

5. Method according to any preceding claim, **characterized** in that the frequency of the to-and-fro mixing movements is about 5-200Hz, preferably about 50Hz.

6. Method according to any preceding claim, **characterized** in that the mixing takes about 0.1-2 seconds, preferably about 0,5 seconds.

7. Method according to any preceding claim, **characterized** in that the to-and-fro mixing movements are provided by the repeated compressions exerted on the pipe (21) which is as an extension to the needle (1) and is of a flexible material, as a result of which compressions the volume of the pipe is changing.

8. Method according to any preceding claim, **characterized** in that the flowing duct (3) of the needle (1) is tapering conically at an angle of about 15-60° towards the opening (4) at the end of the duct.

9. Method according to any preceding claim, **characterized** in that the discharging flow of liquid is directed towards the side walls (7) of the container from openings (4) which are on the needle (1) before its point (5) and are directed to the sides.

10. Method according to any preceding claim, **characterized** in that the needle (1) is lifted during the discharging of the liquid flow, so that the opening(s) (4) follow(s) the rise of the liquid surface (10) in the container (6).

11. A mixing and measuring needle (1) which comprises a tubular shaft (2) and inside it a flowing duct (3), which ends before the point (5) of the needle in at least one

aspiration and discharge opening (4) which is directed aside from the longitudinal axis of the needle, **characterized** in that the point part of the needle has been formed as conically tapering and that in the flowing duct there is a necking (19) extending to said opening, in the length of which necking the flow resistance of the duct is growing at least to tenfold, wherein at discharge stages from the opening according to the vena contracta phenomenon an at first converging and thereafter turbulently divergent liquid flow (20) is discharged.

12. Needle according to claim 11, **characterized** in that the point (5) of the needle is solid.

13. Needle according to claim 11 or 12, **characterized** in that the point part of the needle is tapering off at an angle of between about 15-60°.

14. Needle according to claim 13, **characterized** in that the diameter of the point (5) of the needle is less than 0.3mm.

15. Needle according to claim 12, **characterized** in that the point part of the needle is in the shape of a dagger.

16. Needle according to any of claims 11-15, **characterized** in that the discharge openings (4) have been directed obiquely forward in relation to the longitudinal direction of the needle (1).

17. Needle according to any of claims 11-16, **characterized** in that the flowing duct (3) ends in two or several discharge openings (4) which are at regular intervals on the different sides of the needle (1).

18. A method for manufacturing the mixing and measuring needle (1) according to any of claims 11-17, **characterized** in that the needle (1) is manufactured by electroforming by

electrolytically precipitating around an elongate, needle-like mold (11) a layer (14) of a metal, by removing the mold metal from inside the layer, so that a tubular, metallic needle is obtained having a longitudinal duct (3) inside it, and by drilling on the side of the needle at the end of the duct as the discharge opening (4) of the duct at least one opening (17) which goes through the metal layer.

19. Method according to claim 18, **characterized** in that an excess of the metal is precipitated around the point (12) of the mould (11) and that after the precipitation step the point part (18) of the needle is tooled as conically tapering or in the shape of a dagger, at which stage the extra metal (15) is removed.

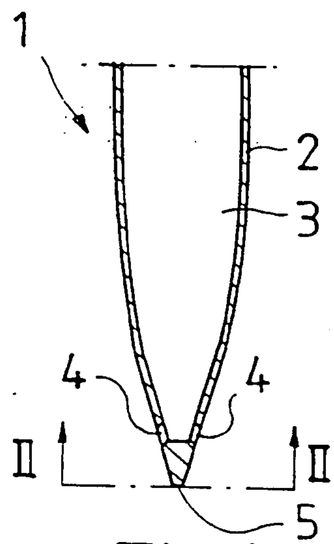

Fig. 1

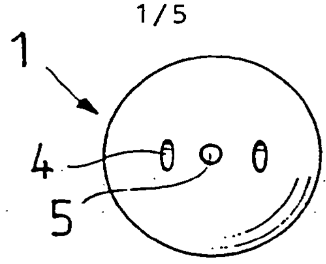

Fig. 2

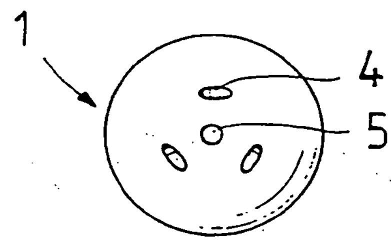

Fig. 3

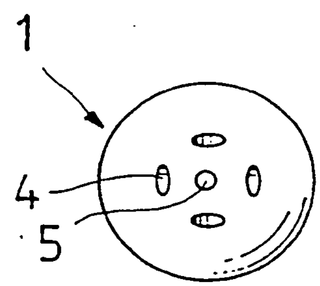

Fig. 4

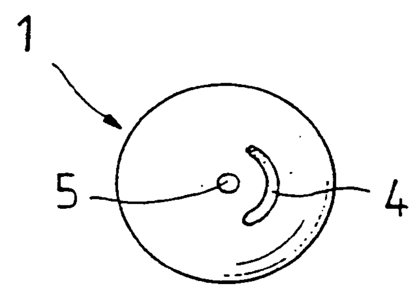

Fig. 5

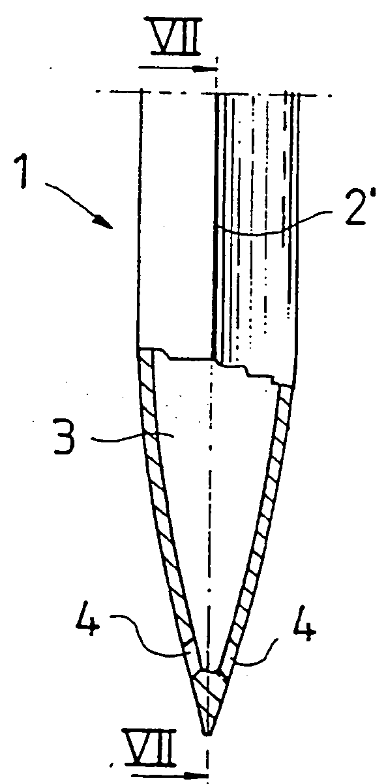

Fig. 6

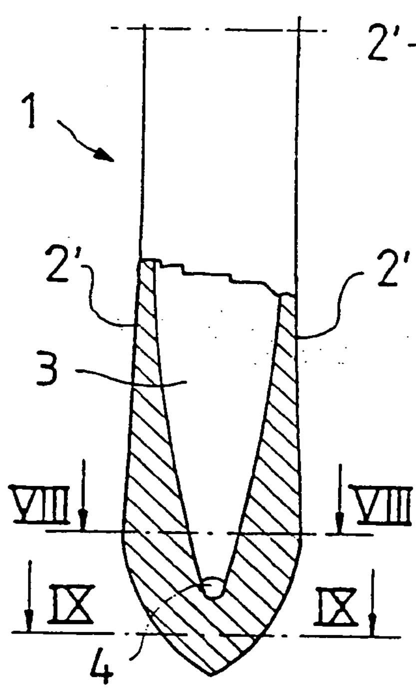

Fig. 7

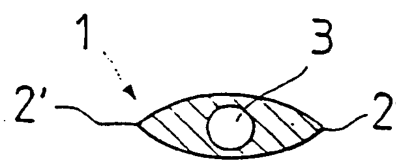

Fig. 8

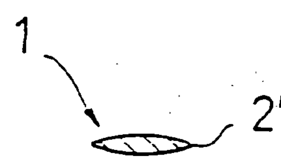

Fig. 9

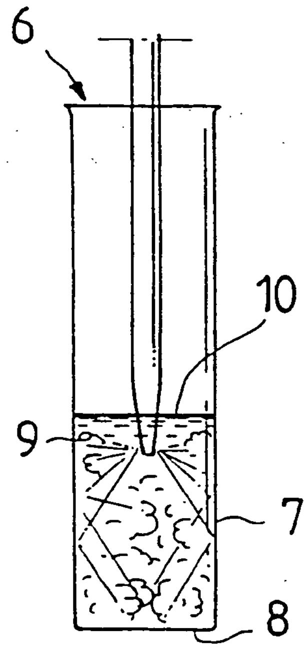

Fig.10

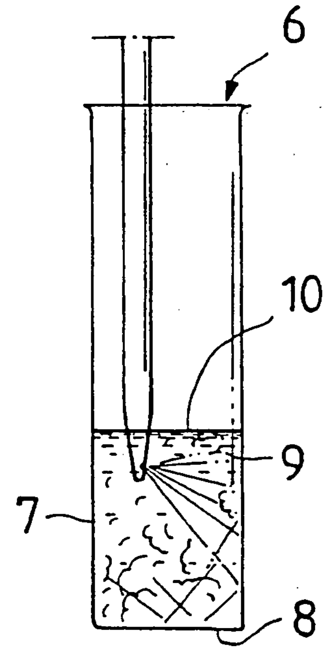

Fig.11

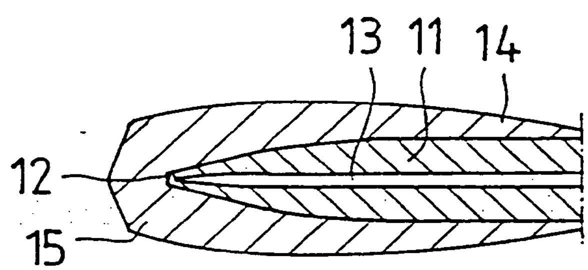

Fig.12

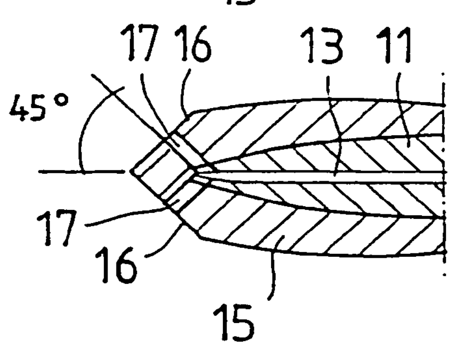

Fig.13

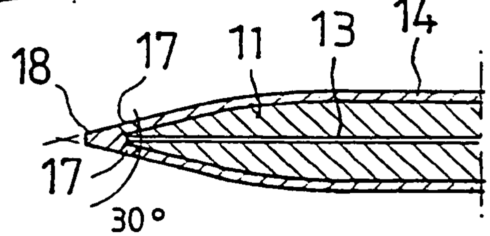

Fig.14

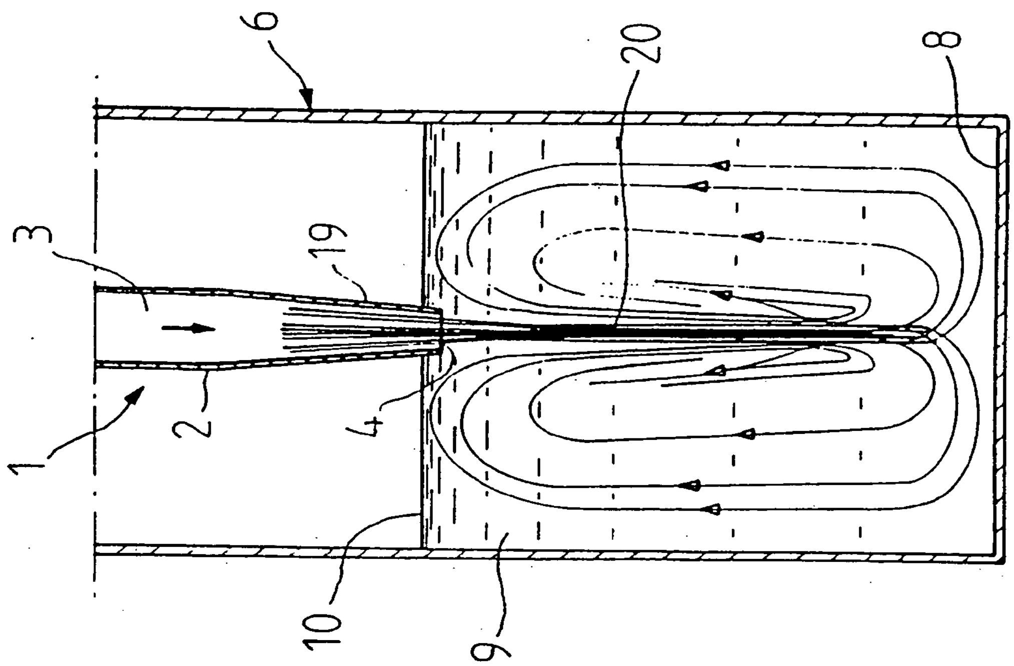

Fig. 16

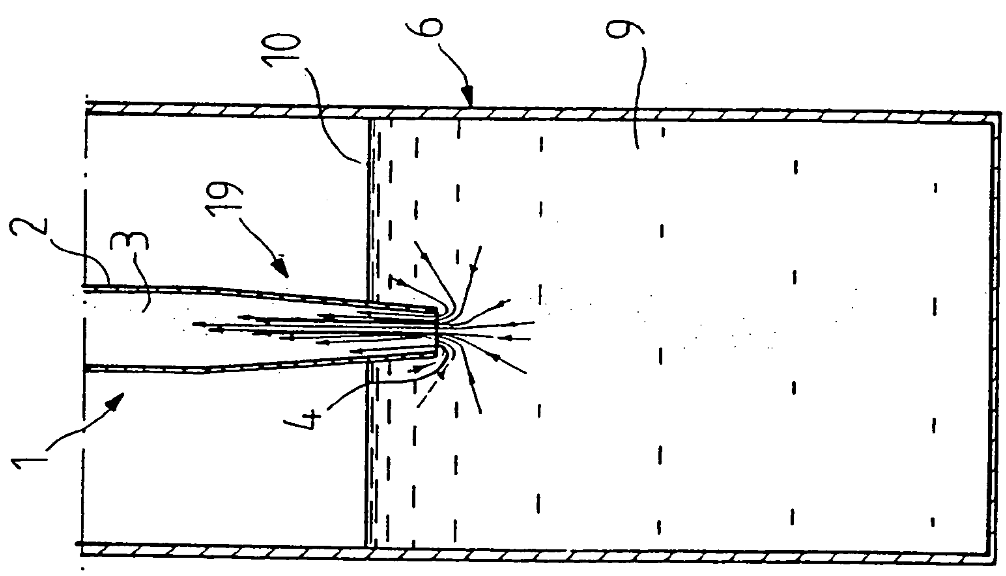

Fig. 15

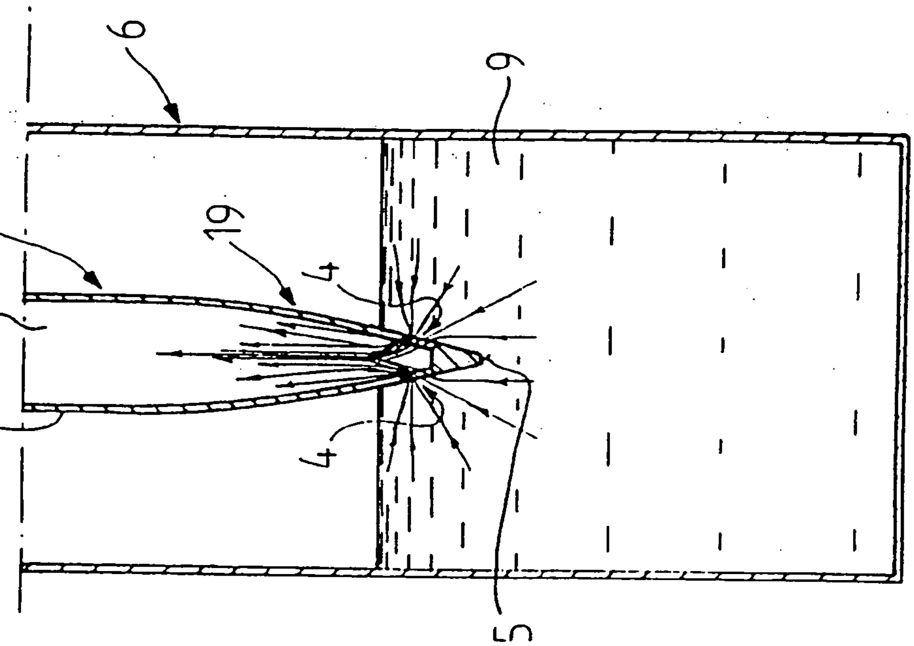

Fig.17

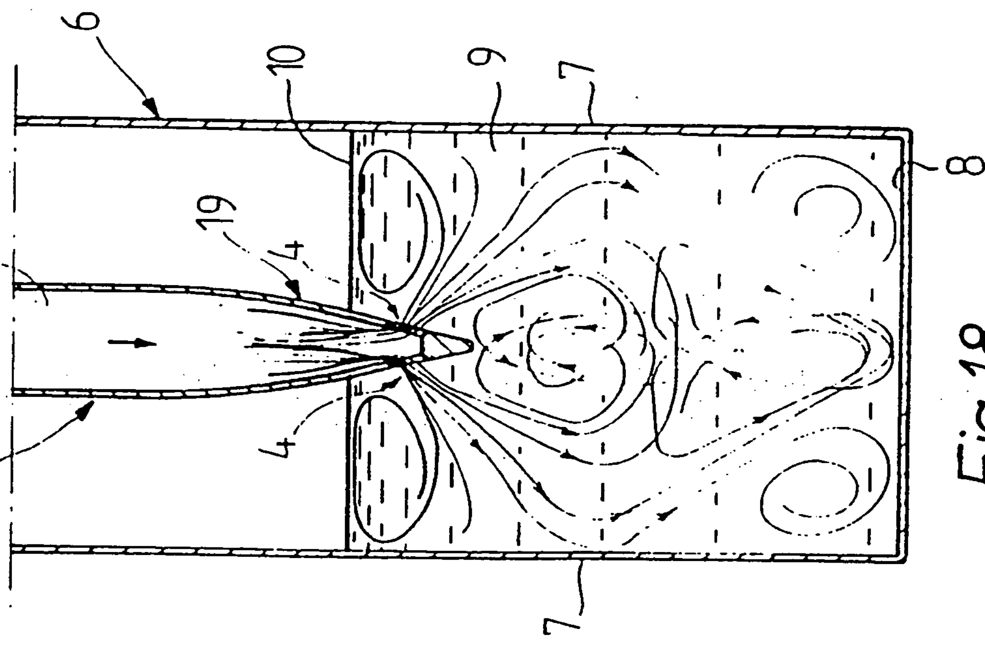

Fig.18

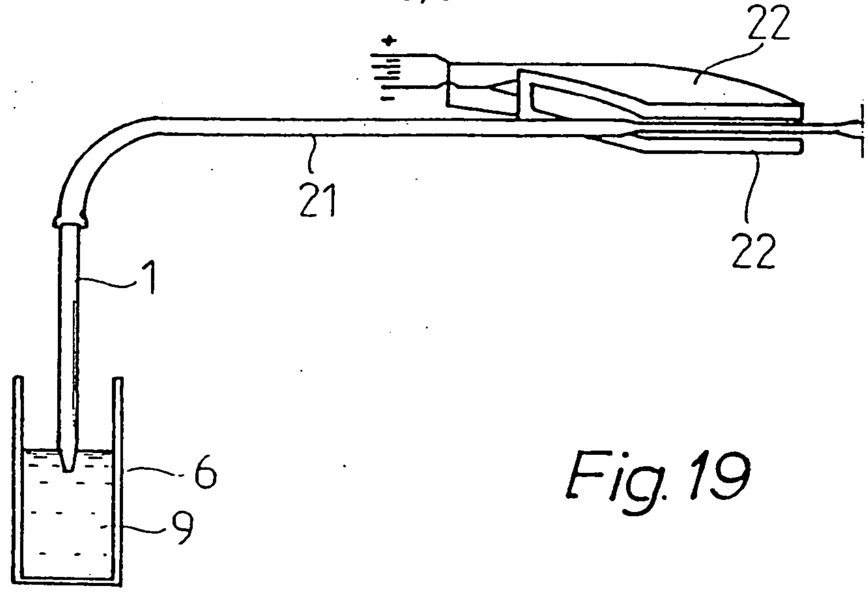

Fig. 19

INTERNATIONAL SEARCH REPORT

International application No.
PCT/FI 93/00263

A. CLASSIFICATION OF SUBJECT MATTER

IPC5: B01L 3/00, B01F 3/08, C25D 1/02

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC5: B01F, B01L, C25D

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

SE,DK,FI,NO classes as above

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	DE, A1, 3115567 (BECKMAN INSTRUMENTS INC.), 8 April 1982 (08.04.82), figures 1,2 --	1-17
A	DE, A1, 2722586 (OLYMPUS OPTICAL CO., LTD.), 24 November 1977 (24.11.77), figure 2 --	11-17
X	GB, A, 2030897 (KERNFORSCHUNGSZENTRUM KARLSRUHE GESELLSCHAFT MIT BESCHRAENKTER HAFTUNG), 16 April 1980 (16.04.80), figure 1, abstract --	18

[X] Further documents are listed in the continuation of Box C. [X] See patent family annex.

* Special categories of cited documents:

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"E" erlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance: the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance: the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
13 Sept 1993	14 -09- 1993
Name and mailing address of the ISA/ Swedish Patent Office Box 5055, S-102 42 STOCKHOLM Facsimile No. +46 8 666 02 86	Authorized officer May Hallne Telephone No. +46 8 782 25 00

INTERNATIONAL SEARCH REPORT

International application No.
PCT/FI 93/00263

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	US, A, 3022697 (E.L. KENT ET AL), 27 February 1962 (27.02.62), column 3, line 28 - line 29 -- --------	18-19

INTERNATIONAL SEARCH REPORT

Information on patent family members

26/08/93

International application No.
PCT/FI 93/00263

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
DE-A1- 3115567	08/04/82	BE-A- 888456	17/08/81
		JP-A- 56158949	08/12/81
		US-A- 4399711	23/08/83
DE-A1- 2722586	24/11/77	NONE	
GB-A- 2030897	16/04/80	DE-A,C- 2828993	17/01/80
		FR-A,B- 2429846	25/01/80
		JP-A- 55038987	18/03/80
		US-A- 4255237	10/03/81
US-A- 3022697	27/02/62	NONE	